CLAIMS

1. An assay method, comprising:

(a) providing a plurality of discrete solid support surface areas,

(b) providing a plurality of different ligands,

(c) defining a first set of different groups of the plurality of ligands,

(d) immobilizing each group of ligands on a different solid support surface area,

(e) providing a plurality of different analytes, each of which is capable of binding to a respective one of the plurality of ligands, at least a major part of the analytes having substantially no cross-reactivity to other ligands,

(f) defining a first set of different groups of the plurality of analytes, each analyte being present in at least one group,

(g) sequentially contacting each group of analytes with the surface areas to bind the analytes in each group to immobilized ligands, and

(h) detecting the interaction of each group of analytes with each group of ligands to determine therefrom the amount of binding of each analyte.

2. The method according to claim 1, wherein at least about 75%, preferably at least about 90% of the analytes, bind specifically to a respective one of the plurality of ligands.

3. The method according to claim 1, wherein each analyte binds specifically to a respective one of the plurality of ligands.

4. The method according to claim 1, wherein none of the different groups of ligands includes all the different ligands.

5. The method according to claim 1, wherein none of the different groups of analytes includes all the different analytes.

6 The method according to claim 1, wherein each ligand is present in at least two different groups of ligands.

7. The method according to claim 1, wherein the groups of ligands and the groups of analytes are defined such that in each group of analytes, each analyte binds specifically to a different one of the different groups of ligands.

8. The method according to claim 1, wherein steps e) to h) in claim 1 are repeated with a second set of different groups of analytes, differently defined than the first set, to determine the possible influence of other analytes on the binding of a specific analyte to a specific ligand.

9. The method according to claim 1, wherein steps b) to h) in claim 1 are repeated with a second set of different groups of ligands, differently defined than the first set, to determine the possible influence of other ligands on the binding of a specific analyte to a specific ligand.

10. The method according to claim 1, wherein each group of analytes contains at least three different analytes.

11. The method according to claim 1, wherein each group of ligands contains at least three different ligands.

12. The method according to claim 7, which comprises providing a plurality of soluble ligands or ligand analogues which bind specifically to respective ones of the analytes, defining different groups of the soluble ligands or ligand analogues such that one ligand or ligand analogue in each group thereof binds specifically to one analyte in each group of analytes, and prior to step g) in claim 1 mixing each group of ligands or ligand analogues with its respective group of analytes.

13. The method according to claim 7, wherein prior to step g) in claim 1, each group of analytes is mixed with binding agents that compete with the analytes for the binding to their respective immobilized ligands.

14. The method according to claim 7, wherein prior to step g) in claim 1, respective specific binding partners to the immobilized ligands are contacted with the different solid support surface areas.

15. The method according to claim 1, wherein after determining the binding of the analytes in a group in step h) in claim 1, the surface areas are contacted with a regeneration solution, and the capability of the regeneration solution to remove each analyte from its ligand is determined.

16. The method according to claim 15, wherein the surface areas subjected to regeneration solution are sequentially contacted with the different groups of analytes to determine any change in binding in relation to that determined in step h) in claim 1.

17. The method according to claim 15, which is repeated with at least one different regeneration solution.

18. The method according to claim 1, wherein the solid support areas are sensing surface areas.

19. The method according to claim 1, wherein the interactions at the surface are monitored in real time.

20. The method according to claim 1, wherein mass changes at the surface areas are detected.

21. The method according to claim 1, wherein the detection is based on evanescent wave sensing.

22. The method according to claim 1, wherein the detection is based on surface plasmon resonance (SPR)

23. The method according to claim 18, wherein the sensing surface areas are provided in at least one flow cell.

24. The method according to claim 1, which comprises determining at least one of ligand immobilization efficiency, analyte concentration, interaction affinity and interaction kinetics.

25. An assay method comprising:

(a) providing a plurality of discrete solid support surface areas,

(b) providing a plurality of different ligands,

(c) defining different groups of the plurality of ligands,

(d) immobilizing each group of ligands on a different solid support surface area,

(e) providing a plurality of different analytes, each analyte having a first binding site and a second binding site, wherein the first binding site of each analyte is capable of binding specifically to a respective one of the plurality of ligands, at least a major part of the first binding sites having substantially no cross-reactivity to other ligands,

(f) contacting each surface area with the analytes to bind the analytes to the immobilized ligands,

(g) providing a plurality of different reagents capable of binding to a respective one of the plurality of analytes at the second binding site thereof, at least a major part of the reagents having substantially no cross-reactivity to other analytes,

(h) defining different groups of the plurality of reagents,

(i) sequentially contacting the surface areas with each group of reagents to bind the reagents in each group to ligand-bound analytes, and

(j) detecting the interaction of each group of reagents with each group of ligand-bound analytes to determine therefrom the amount of binding of each analyte.

26. The method according to claim 25, wherein at least about 90% of the total of analytes and reagents have substantially no cross-reactivity.

27. The method according to claim 25, wherein each analyte is capable of binding specifically to a respective one of the plurality of ligands.

28. The method according to claim 25, wherein each reagent is capable of binding specifically to a respective one of the plurality of analytes.

29. The method according to claim 25, wherein each ligand is present in at least two different groups of ligands.

30. The method according to claim 25, wherein the groups of ligands and the groups of reagents are defined such that in each group of reagents, each reagent binds specifically to a different one of the different groups of immobilized ligands having analytes bound thereto.

31. The method according to claim 25, wherein the solid support areas are sensing surface areas, and the detection is based on evanescent wave sensing.

32. The method according to claim 25, which comprises determining at least one of ligand immobilization efficiency, analyte concentration, interaction affinity and interaction kinetics.

33. An assay method comprising:

(a) providing a plurality of discrete solid support surface areas,

(b) providing a plurality of n different ligands, wherein n is at least 2,

(c) defining different groups of the plurality of n ligands comprising single ligands and combinations of from two to n different ligands,

(d) immobilizing each group of ligands on a different surface area,

(e) sequentially contacting a plurality of n different analytes with each surface area, at least a major part of the analytes being capable of specifically binding to a respective one of the plurality of different ligands, and

(f) detecting the interaction of each analyte with each group of ligands to determine therefrom the amount of ligand-binding of each analyte, and the possible influence of ligand-ligand interaction on the binding of analyte to immobilized ligand.

34. The method according to claim 33, wherein at least about 75%, preferably at least about 90% of the analytes, bind specifically to a respective one of the plurality of ligands.

35. The method according to claim 33, wherein each analyte binds specifically to a respective one of the plurality of ligands.

36. The method according to claim 33, wherein in step e) in claim 33, the surface areas are sequentially contacted with different groups of analytes, comprising single analytes and combinations of from two to n different analytes.

37. The method according to claim 33, wherein the solid support areas are sensing surface areas, and the detection is based on evanescent wave sensing.

38. The method according to claim 33, which comprises determining at least one of ligand immobilization efficiency, analyte concentration, interaction affinity and interaction kinetics.

39. An assay method comprising:

(a) providing a plurality of discrete solid support surface areas,

(b) providing a plurality of n different ligands, wherein n is at least 2,

(c) defining different groups of the plurality of n ligands comprising single ligands and combinations of from two to n different ligands,

(d) immobilizing each group of ligands on a different surface area,

(e) contacting each surface area with a plurality of n analytes to bind them to the ligands, at least a major part of the analytes being capable of binding through a first binding site thereof specifically to a respective one of the plurality of different ligands,

(f) sequentially contacting a plurality of n different reagents with each surface area, at least a major part of the reagents being capable of specifically binding to a respective one of the plurality of different analytes via a second binding site thereof, and

(g) detecting the interaction of each reagent with its ligand-bound analyte to determine therefrom the amount of ligand-binding of each analyte, and the possible influence of ligand-ligand interaction on the binding of analyte to immobilized ligand.

40. The method according to claim 39, wherein at least about 90% of the total of analytes and reagents have substantially no cross-reactivity.

41. The method according to claim 39, wherein each analyte is capable of binding specifically to a respective one of the plurality of ligands.

42. The method according to claim 39, wherein each reagent is capable of binding specifically to a respective one of the plurality of analytes.

43. The method according to claim 39, wherein each ligand is present in at least two different groups of ligands.

44. The method according to claim 39, wherein the groups of ligands and the groups of reagents are defined such that in each group of reagents, each reagent binds specifically to a different one of the different groups of immobilized ligands having analytes bound thereto.

45. The method according to claim 39, wherein in step f) in claim 39, the surface areas are sequentially contacted with different groups of the reagents, comprising single reagents and combinations of from two to n different reagents.

46. The method according to claim 39, wherein the solid support areas are sensing surface areas, and the detection is based on evanescent wave sensing.

47. The method according to claim 39, which comprises determining at least one of ligand immobilization efficiency, analyte concentration, interaction affinity and interaction kinetics.

48. An assay method comprising:

(a) providing a plurality of discrete solid support surface areas,

(b) providing a plurality of different ligands,

(c) defining different groups of the plurality of ligands,

(d) immobilizing each group of ligands on a different solid support surface area,

(e) providing a plurality of different analytes, each of which is capable of binding to a respective one of the plurality of ligands, at least a major part of the analytes having substantially no cross-reactivity to other ligands,

(f) sequentially contacting each analyte with the surface areas to bind the analytes to the immobilized ligands, and

(g) detecting the interaction of analyte with each group of immobilized ligands to determine therefrom the amount of binding of each analyte.

49. The method according to claim 48, wherein at least about 75%, preferably at least about 90% of the analytes, bind specifically to a respective one of the plurality of ligands.

50. The method according to claim 48, wherein each analyte binds specifically to a respective one of the plurality of ligands.

51. The method according to claim 48, wherein the solid support areas are sensing surface areas, and the detection is based on evanescent wave sensing.

52. The method according to claim 48, which comprises determining at least one of ligand immobilization efficiency, analyte concentration, interaction affinity and interaction kinetics.

53. An assay method comprising:

(a) providing a plurality of discrete solid support surface areas,

(b) providing a plurality of different ligands,

(c) defining different groups of the plurality of ligands,

(d) immobilizing each group of ligands on a different solid support surface area,

(e) providing a plurality of different analytes, each analyte having a first binding site and a second binding site, at least a major part of the analytes being capable of binding through the first binding site specifically to a respective one of the different ligands,

(f) contacting each surface area with the analytes to bind the analytes to the immobilized ligands,

(g) providing a plurality of different reagents, at least a major part of the reagents being capable of binding specifically to a respective one of the plurality of analytes at the second binding site thereof,

(h) sequentially contacting each reagent with the surface areas to bind the reagents to the ligand-bound analytes, and

(j) detecting the interaction of each reagent with each surface area to determine therefrom the amount of binding of each analyte.

54. The method according to claim 53, wherein at least about 90% of the total of analytes and reagents have substantially no cross-reactivity.

55. The method according to claim 53, wherein each analyte is capable of binding specifically to a respective one of the plurality of ligands.

56. The method according to claim 53, wherein each reagent is capable of binding specifically to a respective one of the plurality of analytes.

57. The method according to claim 53, wherein each ligand is included in at least two different groups of ligands.

58. The method according to claim 53, wherein the ligands are antibodies and each subgroup of ligands contains different clones of the same antibodies.

59. The method according to claim 53, wherein the solid support areas are sensing surface areas, and the detection is based on evanescent wave sensing.

60. The method according to claim 53, which comprises determining at least one of ligand immobilization efficiency, analyte concentration, interaction affinity and interaction kinetics.

61. A method of determining regeneration conditions for ligand-supporting solid support surfaces, comprising:

(a) providing a plurality of discrete solid support surface areas,

(b) providing a set of a plurality of different ligands,

(c) immobilizing the set of ligands on at least two different surface areas,

(d) sequentially contacting each surface area with a plurality of analytes, at least a major part of the analytes being capable of specifically binding to a respective one of the different ligands,

(e) determining the interaction of each analyte with each surface area having immobilized ligands thereon,

(f) subjecting each surface area having the analytes bound to immobilized ligands to a different regeneration solution,

(g) sequentially contacting the plurality of analytes with each surface area, and

(h) determining for each analyte any change in analyte binding in relation to the binding determined in step e).

62. The method according to claim 61, wherein at least about 75%, preferably at least about 90% of the analytes, bind specifically to a respective one of the plurality of ligands.

63. The method according to claim 61, wherein each analyte binds specifically to a respective one of the plurality of ligands.

64. The method according to claim 61, wherein the solid support areas are sensing surface areas, and the detection is based on evanescent wave sensing.